The immunology of the Periodic Fever, Aphthous stomatitis, Pharyngitis and cervical Adenitis syndrome; what can the tonsils reveal. A literature review.

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Abstract

Objectives

Tonsillectomy (TE) or adenotonsillectomy (ATE) may have a beneficial effect on the clinical course in children with the periodic fever, aphthous stomatitis, pharyngitis and cervical adenitis (PFAPA) syndrome. However, an immunological reason for this effect remains unknown. This literature review summarizes the current knowledge regarding the immunological role of the tonsils in the PFAPA syndrome.

Methods

We searched PubMed, Medline, EMBASE and Cochrane for papers written in English dated from 1 January 1987 to 30 April 2019. The search included all studies reporting histological, immunological or microbiological workup of tonsil specimens from children aged 0 to 18 years with PFAPA.

Results

Thirteen articles reported histological, immunological or microbiological workup of tonsil specimens in children with PFAPA. The histology of tonsil specimens from children with PFAPA displayed chronic tonsillar inflammation with lymphoid hyperplasia. No uniform immunological pattern was identified, but some studies found fewer B-lymphocytes and smaller germinal centers in PFAPA compared to controls. A difference in tonsillar microbiota between PFAPA and controls was found in one study.

Conclusion

A uniform immunological or microbiological pattern explaining the clinical effect of TE in children with PFAPA has not been revealed. Future targeted immunological studies of tonsils in PFAPA patients could possibly illuminate the understanding of the immunology in this disease.

Keywords:

PFAPA, Tonsils, Periodic Fever

Conflict of Interest: The authors declare that they have no conflict of interest.

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

1. Introduction

The periodic fever, aphthous stomatitis, pharyngitis and cervical adenitis (PFAPA) syndrome is the most common paediatric periodic fever syndrome [1, 2], with a cumulative incidence of 2.2 per 10,000 children up to the age of 5 years in a Nordic population [2]. The hallmarks of the disease are short, regularly occurring episodes of high fever accompanied by at least one of the following major symptoms: pharyngitis, cervical adenitis and aphthous stomatitis [3-5]. Episodes of PFAPA often start during the first few years of life and often spontaneously resolve during late childhood [2, 6]. The criteria for establishing a PFAPA diagnosis was suggested by Thomas et al. in 1999 and have been widely used in international studies, however, there is no established international consensus regarding the definition of PFAPA today [7, 8].

A dysregulated interleukin (IL)-1 response may play a part in the etiology of the disease [9, 10], and PFAPA is currently regarded as an autoinflammatory disease [11]. Unlike the classical hereditary periodic fever syndromes including familial Mediterranean fever, mevalonate kinase deficiency and TNF-receptor associated periodic syndrome, PFAPA has not been defined genetically. Several candidate genes have been proposed as contributing factors to the etiology of PFAPA, and in addition some studies show familiar clustering of the disease. A polygenic basis for the disease is likely, but further studies are needed to elucidate this. As recently reviewed, an environmental trigger causing immunological reactions with febrile flares in a genetic predisposed host seems the best founded model for the disease today [12].

Since PFAPA was first described as a clinical entity by Marshall et al. in 1987 [13], a beneficial effect of tonsillectomy (TE) or adenotonsillectomy (ATE) has been reported in several case series [14] and two randomized controlled trials of children with PFAPA [15, 16]. Both a Cochrane review and a recent review of all case series conclude that TE or ATE

may have a curative effect on children with PFAPA, but the evidence is of moderate quality [14, 17].

The palatine tonsils (referred to as the tonsils in this paper), together with the adenoids and the lingual tonsils constitute a large part of what is called Waldeyer's ring. This lymphoid tissue is strategically located in the aerodigestive tract exposed to airborne and alimentary antigens [18]. The reason why TE or ATE may have a beneficial effect in children with PFAPA is not known. **Some studies have explored different immunological aspects of the tonsils and the tonsillar microbiota in children with PFAPA.** The aim of this article was to summarize the results of all studies reporting histological, immunological and microbiological findings in tonsils from these children.

2. **Methods**

A systematic search of the PubMed, Medline, EMBASE and Cochrane databases was performed up to April 30th 2019 using the keywords "Marshall Syndrome", "PFAPA" and "Periodic Fever, Aphthous stomatitis, Pharyngitis and cervical Adenitis". Papers in English language published between 1 January 1987 and 30 April 2019 were checked for relevance. The references in the relevant papers were also reviewed to identify any articles not found in the systematic search. Papers were included if they reported histological, immunological or microbiological studies of tonsil specimens from children aged 0 to 18 years diagnosed with PFAPA. **The diagnostic criteria for the diagnosis in each study were noted; if they were according to Marshall [13], Thomas [7] or adapted criteria not clearly based on either of the two. The diagnostic criteria according to Marshall and Thomas are presented in Table 1.**

3. **Results**

The search retrieved 13 articles reporting microbiological, histological or immunological workup of tonsil specimens in children with PFAPA (Figure 1). **Two articles included only one and two patients with PFAPA, and were therefore not included in the further presentation [19, 20]. Data from the remaining 11 articles including four or more children are summarized in Table 2.**

3.1 *Histological and immunological evaluation of tonsils in PFAPA*

The histology of tonsils from children with PFAPA has been evaluated in eight studies [21-28]. All studies reported similar findings, with tonsil specimens showing signs of chronic tonsillar inflammation with lymphoid hyperplasia. Four of these studies compared the histologic findings with a control group [24-26, 28]. With the exception of one study presenting differences in measurements of germinal centers and epithelium width (presented in detail below) [28], the studies showed no difference between the children with PFAPA and the controls.

In six studies, the tonsils were characterized further by immunhistochemical staining for various cell types and protein expression and/or gene expression studies [23-25, 28-30].

Peridis et al. reported preservation of normal tonsillar architecture after immunohistochemically staining of T and B lymphocytes, using markers for cluster of differentiation (CD) 3 and CD20, respectively [23].

Dytrych et al. examined manually dissociated tonsil tissue from children with PFAPA compared to controls with obstructive sleep apnoea syndrome. They found a lower percentage of B lymphocytes (CD19+) and an increased proportion of cytotoxic T cells (CD8+). Additionally, some subsets of lymphocytes (translational B cells, naïve stages of CD4+ and CD8+ cells with a low expression of the PD-1 molecule and a high number of T-cell receptor excision circles) were found to be increased in tonsil tissue from children with PFAPA.

Polymerase chain reaction (PCR) studies for rearrangement of immunoglobulin and T cell receptors did not show clonal or oligoclonal expansion. The expression of the chemokine CXCL10, CXCL9 and CCL19 genes were significantly higher in the tonsils of children with PFAPA syndrome. The authors interpreted that their results may indicate that the differences in the T-cell compartment observed in tonsils and not in blood could partly explain the effect of tonsillectomy seen in children with PFAPA, and further that the altered gene expression of T cell chemoattractants in tonsils may be a precipitating cause of PFAPA [24].

Førsvoll et al. compared tonsils from children with PFAPA and controls with tonsillar hypertrophy. They found a decreased number of cytotoxic T cells (CD8+) in tonsillar germinal centers in children with PFAPA compared to controls, but they found no differences for the other cell types studied (CD3+, CD4+, CD15+, CD20+, CD45+, CD57+ and CD163+). The authors interpreted that their results may indicate that the regulation of CD8+ T lymphocytes homing to lymphoid follicles in tonsils is linked to the etiology of PFAPA [25].

Manthiram et al. measured the size of germinal centers, mantel width, interfollicular distance ant the width of the squamous epithelium and performed immunohistochemical staining in cases and controls with obstructive sleep apnea. The two groups were clearly defined as a polysomnography was performed in all participating children. The average germinal center area was smaller and the average squamous epithelium width was wider in children with PFAPA compared to children with obstructive sleep apnea. Mantle width and interfollicular distance were not significantly different. In children with PFAPA the size of the tonsil germinal centers were larger with longer time from the last febrile episode. Positive staining for the CD markers (CD1a, CD3, CD4, CD8, CD11c, CD14, CD20 CD56, CD68), Foxp3, TUBB2A and IL-6 and IL-1RA expression were not significantly different in children with PFAPA and controls in any of the major histologic compartments (crypt, germinal

center, and surface squamous epithelium). The authors interpreted that their results may indicate that tonsils from children with PFAPA change histologically over time and that germinal centers may shrink around the time of PFAPA flares [28].

Gazi et al. compared tonsillar expression levels of antimicrobial peptides, namely human beta-defensin 1 and 2, cathelicidin, ribonuclease-7, and liver expressed antimicrobial peptide-1, between children with PFAPA and controls with group A beta-hemolytic streptococcal recurrent tonsillitis. They found no difference in the overall expression of antimicrobial peptides between cases and controls. However, they found a different pattern of localization of human beta-defensin 1 when comparing the two groups. The authors interpreted that their results may indicate that PFAPA patients have altered antimicrobial peptide expression pattern compared to normal tonsils [30].

Valenzuela et al. compared tonsils from children with PFAPA with tonsils from controls operated on due to hypertrophic tonsils. They reported lower gene expression for IL-4 in tonsils from children with PFAPA compared to controls, but otherwise they observed no differences in gene expression for IL-1β, IL-17, tumor necrosis factor-α, transforming growth factor-β and interferon-γ. The authors interpreted that their results may indicate that an inhibition of Th2 responses in tonsils is linked to the pathogenesis of PFAPA [29].

3.2 *Microbiological evaluation of tonsils*

In two studies published by a group in Finland where the data apparently is collected from the same group of patients, the tonsillar microbiota in children with PFAPA was explored using bacterial and yeast cultures, virus PCR, electron microscopy of biofilm and bacterial DNA extraction [26, 31]. The culture and PCR tests showed that *Candida albicans* was more common in the PFAPA patients than in the controls, and *Staphylococcus aureus*, Varicella zoster and Herpes simplex viruses occurred less often in the children with PFAPA than in the controls. In addition, biofilm was more frequently present in the tonsils from the children with PFAPA. When using sequencing technology, the study group also found some significant differences in the microbiota of the tonsils from the children with PFAPA compared to the controls. Cyanobacteria were more common in children with PFAPA than in the controls. Streptococci, although present in all samples of both groups, showed lower relative abundance in children with PFAPA than in the controls. The authors interpreted that their results may indicate that the tonsillar microbiota play a role in triggering the inflammatory process causing PFAPA, and that removal of this triggering microbial factor in part explain why TE is effective in children with PFAPA [26, 31].

Furthermore, Dytrych et al. found DNA from the Epstein-Barr virus, Human Herpervirus-6 or adenovirus in 7 of 10 tonsil specimens from children with PFAPA, but also in seven of nine controls [24].

4. **Discussion**

Several studies have indicated that a dysregulated immune system may play a part in the aetiology of PFAPA [9, 10, 32-35]. In 2011, Stojanov et al. proposed a model for PFAPA where a microbial trigger initiates a cascade leading to the febrile attacks. They suggested that an immunologically immature host or a host with an inherited or acquired immune

abnormality plays a permissive role [9]. The observation that removal of the tonsils, a minor part of the adaptive immune system, may be curative for a disease that is possibly caused by a generally dysregulated immune system is puzzling. **Although both tonsillectomy and adenoidectomy were done in some cases, immunological workup was described only on tonsil specimens.** The histological and microbiological studies of tonsils in children with PFAPA published so far reveals no clear answer to this puzzle.

4.1 *Histological and immunological evaluation*

The histology of tonsil specimens from children with PFAPA reveals chronic tonsillar inflammation with lymphoid hyperplasia. Together with the evident pharyngitis present in the majority of patients during febrile attacks, this clearly shows that the tonsils are an immunological active organ in PFAPA. However, the signs of inflammation were similar in controls, and may therefore be considered as inflammatory changes not specific for PFAPA.

The existing studies on the immunological aspects of tonsils in PFAPA differ in design and focus, and they are not easily comparable. There is no common thread in their results, and there is not a clear answer regarding how the immunology of the tonsils contributes to the pathogenesis of PFAPA or why TE may be beneficial. However, the results point out some apparent interesting aspects of tonsillar immunology, and may indicate areas that should be elucidated in future studies. The immaturity of polyclonal lymphocytes with increased T-cell receptor excision circles content and higher expression of chemoattractants may be interpreted as the influx of thymal recruits, reactive to the febrile episode [24]. Dytriych et al. (2015) and Manthiram et al. (2018) found fewer B-lymphocytes and smaller germinal centers in tonsils indicating some regulatory disturbances, possibly by T cells. Indeed, Førsvoll et al. (2013) demonstrated significantly fewer follicular CD8+ T cells in tonsils of PFAPA children. Future studies using quantitative multiplex immunohistochemistry

could reveal cell subtypes in more clarifying detail, and can even be performed in archival material [36].

There is a paucity of publications focusing on the cyclic, infradian, nature of PFAPA. Clock-related genes and their relation to immunity are well described (reviewed in [37]), but most publications on clock genes relate to circadian and not the infradian rhythms as seen in PFAPA. Yet, there are rhythmic fluctuations of inflammatory pathways and clock genes are associated with increased cellular oxygen concentration [38]. Probing into clock genes in tonsillar tissues may therefore be of interest. Another possible approach may be to probe expressed genes of the inflammasomes in PFAPA tonsils [39].

TE or ATE in children with PFAPA are performed during the afebrile interval, and this may limit the information provided when studying the immunology of the tonsils, as important immunological aspects of PFAPA may be absent outside the febrile intervals. This may be illustrated by Manthiram et al.'s finding of altered tonsil germinal centers size with longer time from the last febrile episode [28]. Tonsil specimens are not available from healthy controls, and results found when comparing tonsils from children with PFAPA and from children with other diseases can be difficult to interpret.

4.2 *Microbiological evaluation of tonsils*

Some differences in the presence of *Candida albicans* and some common viruses and bacteria were found in the Finnish studies comparing biofilm samples from children with PFAPA and a control group [26, 31]. This could indicate that children with PFAPA have an altered tonsillar microbiological milieu. However, the varying degrees of biofilm, bacteria and fungi present in the upper airways may also have been caused by differences in the use of antibiotics prior to surgery in children with and without PFAPA.

The lack of a clear clinical definition for PFAPA and inclusion criteria for patients could also bias studies regarding the possible role of microbiological agents in PFAPA. Padeh et al. diagnosed PFAPA only in children with no effect of antibiotic treatment, either as a treatment during febrile episodes or as a prophylaxis, or when they had negative throat cultures during the attacks [3]. Thomas et al. and Tasher et al. had a less systematic approach, but their findings are comparable, with no effect from antibiotics in 92% and 98% of children, respectively [5, 7].

4.3 *Limitation of the studies*

PFAPA is diagnosed using clinical criteria, and although the criteria presented by Thomas et al. in 1999 are the most commonly used in studies, there is no international consensus. This may cause heterogeneity in study populations and thus in results of immunological studies. Recently, the Eurofever network has proposed classification criteria for PFAPA intended for clinical, epidemiological and translational studies [40]. These criteria are not meant to be diagnostic criteria for clinical practice, but as inclusion criteria in studies. The authors argue that this will facilitate the accurate identification of PFAPA and differentiating it from other autoinflammatory diseases.

The studies performed on children with PFAPA are performed on study populations ranging from 4 to 31 cases with 0 to 24 controls. There is a risk of finding a false positive or a false negative results when a high number of comparisons are made on relatively few patients and controls [41]. This may be the cause of the lack of reproducibility for some of the results seen in the different studies of tonsils in PFAPA. Collecting children in order to reach a sufficient sample size for a tonsillectomy-PFAPA-study is time consuming, and to help this problem international multi-center collaboration should be considered when planning future studies.

5 Conclusions

Only a few studies have explored immunological and microbiological aspects of tonsils in children with PFAPA, suggesting a role for the tonsils in the pathophysiology of PFAPA, but no common immunological explanation for the possible effect of TE has been found. More and thorough immunological studies could possibly substantiate the possible effect of TE or ATE in PFAPA and illuminate the understanding of the immunology in this disease.

List of abbreviations:

ATE: Adenotonsillectomy

CD: Cluster of differentiation

IL: Interleukin

PCR: Polymerase chain reaction

PFAPA: Periodic Fever, Aphthous stomatitis, Pharyngitis and cervical Adenitis

TE: Tonsillectomy

Declarations

Authors' contributions

The authors have contributed equally to the manuscript. JF performed the literature search. KØ wrote the first draft of the manuscript. All authors revised the manuscript and prepared it for publication.

References

[1] Masters SL, Simon A, Aksentijevich I, Kastner DL, Horror autoinflammaticus: the molecular pathophysiology of autoinflammatory disease (*), Annu Rev Immunol. 27 (2009) 621-668.
[2] Forsvoll J, Kristoffersen EK, Oymar K, Incidence, clinical characteristics and outcome in Norwegian children with periodic fever, aphthous stomatitis, pharyngitis and cervical adenitis syndrome; a population-based study, Acta Paediatr. 102 (2013) 187-192.
[3] Padeh S, Brezniak N, Zemer D, Pras E, Livneh A, Langevitz P, Migdal A, Pras M, Passwell JH, Periodic fever, aphthous stomatitis, pharyngitis, and adenopathy syndrome: Clinical characteristics and outcome, Journal of Pediatrics. 135 (1999) 98-101.
[4] Thomas KT, Feder HM, Jr., Lawton AR, Edwards KM, Periodic fever syndrome in children, Journal of Pediatrics. 135 (1999) 15-21.
[5] Tasher D, Somekh E, Dalal I, PFAPA syndrome: new clinical aspects disclosed, Archives of Disease in Childhood. 91 (2006) 981-984.
[6] Wurster VM, Carlucci JG, Feder HM, Jr., Edwards KM, Long-term follow-up of children with periodic fever, aphthous stomatitis, pharyngitis, and cervical adenitis syndrome, Journal of Pediatrics. 159 (2011) 958-964.
[7] Thomas KT, Feder Jr HM, Lawton AR, Edwards KM, Periodic fever syndrome in children, Journal of Pediatrics. 135 (1999) 15-21.
[8] Hofer M, Pillet P, Cochard MM, Berg S, Krol P, Kone-Paut I, Rigante D, Hentgen V, Anton J, Brik R, Neven B, Touitou I, Kaiser D, Duquesne A, Wouters C, Gattorno M, International periodic fever, aphthous stomatitis, pharyngitis, cervical adenitis syndrome cohort: description of distinct phenotypes in 301 patients, Rheumatology (Oxford). 53 (2014) 1125-1129.
[9] Stojanov S, Lapidus S, Chitkara P, Feder H, Salazar JC, Fleisher TA, Brown MR, Edwards KM, Ward MM, Colbert RA, Sun HW, Wood GM, Barham BK, Jones A, Aksentijevich I, Goldbach-Mansky R, Athreya B, Barron KS, Kastner DL, Periodic fever, aphthous stomatitis, pharyngitis, and adenitis (PFAPA) is a disorder of innate immunity and Th1 activation responsive to IL-1 blockade, Proc Natl Acad Sci U S A. 108 (2011) 7148-7153.
[10] Kolly L, Busso N, Von Scheven-Gete A, Bagnoud N, Moix I, Holzinger D, Simon G, Ives A, Guarda G, So A, Morris MA, Hofer M, Periodic fever, aphthous stomatitis, pharyngitis, cervical adenitis syndrome is linked to dysregulated monocyte IL-1beta production, Journal of Allergy and Clinical Immunology. 131 (2013) 1635-1643.
[11] Wekell P, Karlsson A, Berg S, Fasth A, Review of autoinflammatory diseases, with a special focus on periodic fever, aphthous stomatitis, pharyngitis and cervical adenitis syndrome, Acta Paediatr. 105 (2016) 1140-1151.
[12] Wekell P, Periodic fever, aphthous stomatitis, pharyngitis, and cervical adenitis syndrome - PFAPA syndrome, Presse Med. 48 (2019) e77-e87.
[13] Marshall GS, Edwards KM, Butler J, Lawton AR, Syndrome of periodic fever, pharyngitis, and aphthous stomatitis, Journal of Pediatrics. 110 (1987) 43-46.
[14] Forsvoll J, Oymar K, The role of tonsillectomy in the Periodic Fever, Aphthous stomatitis, Pharyngitis and cervical Adenitis syndrome; a literature review, BMC Ear Nose Throat Disord. 18 (2018) 3.
[15] Renko M, Salo E, Putto-Laurila A, Saxen H, Mattila PS, Luotonen J, Ruuskanen O, Uhari M, A Randomized, Controlled Trial of Tonsillectomy in Periodic Fever, Aphthous Stomatitis, Pharyngitis, and Adenitis Syndrome, Journal of Pediatrics. 151 (2007) 289-292.

[16] Garavello W, Romagnoli M, Gaini RM, Effectiveness of Adenotonsillectomy in PFAPA Syndrome: A Randomized Study, Journal of Pediatrics. 155 (2009) 250-253.
[17] Burton MJ, Pollard AJ, Ramsden JD, Chong LY, Venekamp RP, Tonsillectomy for periodic fever, aphthous stomatitis, pharyngitis and cervical adenitis syndrome (PFAPA), Cochrane Database Syst Rev. 9 (2014) CD008669.
[18] Brandtzaeg P, Immune functions of nasopharyngeal lymphoid tissue, Advances in oto-rhino-laryngology. 72 (2011) 20-24.
[19] Chen K, Xu W, Wilson M, He B, Miller NW, Bengten E, Edholm ES, Santini PA, Rath P, Chiu A, Cattalini M, Litzman J, J BB, Huang B, Meini A, Riesbeck K, Cunningham-Rundles C, Plebani A, Cerutti A, Immunoglobulin D enhances immune surveillance by activating antimicrobial, proinflammatory and B cell-stimulating programs in basophils, Nature immunology. 10 (2009) 889-898.
[20] K. T, T. S, Y. I, H. Y, K. U, Tonsillectomy as treatment for periodic fever, aphthous stomatitis, pharyngitis, and cervical adenitis syndrome., Journal of Nepal Paediatric Society. 37 (2017) 101-103.
[21] Abramson JS, Givner LB, Thompson JN, Possible role of tonsillectomy and adenoidectomy in children with recurrent fever and tonsillopharyngitis, Pediatr Infect Dis J. 8 (1989) 119-120.
[22] Berlucchi M, Meini A, Plebani A, Bonvini MG, Lombardi D, Nicolai P, Update on treatment of Marshall's syndrome (PFAPA syndrome): Report of five cases with review of the literature, Annals of Otology, Rhinology and Laryngology. 112 (2003) 365-369.
[23] Peridis S, Koudoumnakis E, Theodoridis A, Stefanaki K, Helmis G, Houlakis M, Surgical outcomes and histology findings after tonsillectomy in children with periodic fever, aphthous stomatitis, pharyngitis, and cervical adenitis syndrome, Am J Otolaryngol. 31 (2010) 472-475.
[24] Dytrych P, Krol P, Kotrova M, Kuzilkova D, Hubacek P, Krol L, Katra R, Hrusak O, Kabelka Z, Dolezalova P, Kalina T, Fronkova E, Polyclonal, newly derived T cells with low expression of inhibitory molecule PD-1 in tonsils define the phenotype of lymphocytes in children with Periodic Fever, Aphtous Stomatitis, Pharyngitis and Adenitis (PFAPA) syndrome, Molecular immunology. 65 (2015) 139-147.
[25] Forsvoll J, Janssen EAM, Moller I, Wathne N, Skaland I, Klos J, Kristoffersen EK, Oymar K, Reduced Number of CD8+ Cells in Tonsillar Germinal Centres in Children with the Periodic Fever, Aphthous Stomatitis, Pharyngitis and Cervical Adenitis Syndrome, Scandinavian journal of immunology. 82 (2015) 76-83.
[26] Lantto U, Koivunen P, Tapiainen T, Glumoff V, Hirvikoski P, Uhari M, Renko M, Microbes of the tonsils in PFAPA (Periodic Fever, Aphtous stomatitis, Pharyngitis and Adenitis) syndrome - A possible trigger of febrile episodes, APMIS : acta pathologica, microbiologica, et immunologica Scandinavica. 123 (2015) 523-529.
[27] Batu ED, Kara Eroglu F, Tsoukas P, Hausmann JS, Bilginer Y, Kenna MA, Licameli GR, Fuhlbrigge RC, Ozen S, Dedeoglu F, Periodic Fever, Aphthosis, Pharyngitis, and Adenitis Syndrome: Analysis of Patients From Two Geographic Areas, Arthritis care & research. 68 (2016) 1859-1865.
[28] Manthiram K, Correa H, Boyd K, Roland J, Edwards K, Unique histologic features of tonsils from patients with periodic fever, aphthous stomatitis, pharyngitis, and cervical adenitis (PFAPA) syndrome, Clin Rheumatol. 37 (2018) 1309-1317.
[29] Valenzuela PM, Araya A, Perez CI, Maul X, Serrano C, Beltran C, Harris PR, Talesnik E, Profile of inflammatory mediators in tonsils of patients with periodic fever, aphthous stomatitis, pharyngitis, and cervical adenitis (PFAPA) syndrome, Clin Rheumatol. 32 (2013) 1743-1749.

[30] Gazi U, Agada ME, Ozkayalar H, Dalkan C, Sanlidag B, Safak MA, Mocan G, Bahceciler NO, Tonsillar antimicrobial peptide (AMP) expression profiles of periodic fever, aphthous stomatitis, pharyngitis, cervical adenitis (PFAPA) patients, Int J Pediatr Otorhinolaryngol. 110 (2018) 100-104.
[31] Tejesvi MV, Uhari M, Tapiainen T, Pirttila AM, Suokas M, Lantto U, Koivunen P, Renko M, Tonsillar microbiota in children with PFAPA (periodic fever, aphthous stomatitis, pharyngitis, and adenitis) syndrome, Eur J Clin Microbiol Infect Dis. (2016).
[32] Stojanov S, Hoffmann F, Kery A, Renner ED, Hartl D, Lohse P, Huss K, Fraunberger P, Malley JD, Zellerer S, Albert MH, Belohradsky BH, Cytokine profile in PFAPA syndrome suggests continuous inflammation and reduced anti-inflammatory response, Eur Cytokine Netw. 17 (2006) 90-97.
[33] Brown KL, Wekell P, Osla V, Sundqvist M, Savman K, Fasth A, Karlsson A, Berg S, Profile of blood cells and inflammatory mediators in periodic fever, aphthous stomatitis, pharyngitis and adenitis (PFAPA) syndrome, BMC Pediatr. 10 (2010) 65.
[34] Forsvoll J, Kristoffersen EK, Oymar K, Elevated levels of CXCL10 in the Periodic Fever, Aphthous stomatitis, Pharyngitis and cervical Adenitis syndrome (PFAPA) during and between febrile episodes; an indication of a persistent activation of the innate immune system, Pediatric rheumatology online journal. 11 (2013) 38.
[35] Cheung MS, Theodoropoulou K, Lugrin J, Martinon F, Busso N, Hofer M, Periodic Fever with Aphthous Stomatitis, Pharyngitis, and Cervical Adenitis Syndrome Is Associated with a CARD8 Variant Unable To Bind the NLRP3 Inflammasome, J Immunol. 198 (2017) 2063-2069.
[36] Stack EC, Wang C, Roman KA, Hoyt CC, Multiplexed immunohistochemistry, imaging, and quantitation: a review, with an assessment of Tyramide signal amplification, multispectral imaging and multiplex analysis, Methods. 70 (2014) 46-58.
[37] Scheiermann C, Gibbs J, Ince L, Loudon A, Clocking in to immunity, Nature reviews. Immunology. 18 (2018) 423-437.
[38] Lagishetty V, Parthasarathy PT, Phillips O, Fukumoto J, Cho Y, Fukumoto I, Bao H, Cox R, Jr., Galam L, Lockey RF, Kolliputi N, Dysregulation of CLOCK gene expression in hyperoxia-induced lung injury, Am J Physiol Cell Physiol. 306 (2014) C999-C1007.
[39] Armani M, Tangrea M, Yang B, Rosenberg A, Ylaya K, Morris J, Rodriguez-Canales J, Hanson J, Shapiro B, Emmert-Buck MR, Smela E, Hewitt SM, Multiplex quantitative measurement of mRNAs from fixed tissue microarray sections, Applied immunohistochemistry & molecular morphology : AIMM / official publication of the Society for Applied Immunohistochemistry. 22 (2014) 323-330.
[40] Gattorno M, Hofer M, Federici S, Vanoni F, Bovis F, Aksentijevich I, Anton J, Arostegui JI, Barron K, Ben-Cherit E, Brogan PA, Cantarini L, Ceccherini I, De Benedetti F, Dedeoglu F, Demirkaya E, Frenkel J, Goldbach-Mansky R, Gul A, Hentgen V, Hoffman H, Kallinich T, Kone-Paut I, Kuemmerle-Deschner J, Lachmann HJ, Laxer RM, Livneh A, Obici L, Ozen S, Rowczenio D, Russo R, Shinar Y, Simon A, Toplak N, Touitou I, Uziel Y, van Gijn M, Foell D, Garassino C, Kastner D, Martini A, Sormani MP, Ruperto N, Classification criteria for autoinflammatory recurrent fevers, Ann Rheum Dis. (2019).
[41] Sedgwick P, Multiple hypothesis testing and Bonferroni's correction, BMJ. 349 (2014) g6284.